A
Chemical
LIGHT
METER

for Forest
Research

**David A. Marquis
& George Yelenosky**

THE AUTHORS —

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To Measure Light

LIGHT is one of the most important factors that influence the growth and development of forest trees. Not only is light necessary for the basic process of photosynthesis; light also regulates and modifies other factors of the environment such as temperature and moisture. Yet despite the importance of light, few investigators have succeeded in measuring it adequately in the forest. The primary reason for this failure is the lack of suitable instruments or methods for measuring and summating light over a period of time.

The problem usually is to compare light intensities among several locations. To do this, one should simultaneously record the

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total amount of light that reaches each location during a period of time — say one day. In this way, differences due to the sun's position, and to the shadows cast by clouds and by the forest canopy, are expressed in the light measurements.

Exposure meters or illumination meters, which measure light intensity at a given moment, yield data of very limited utility unless a large number of readings, taken over a period of time, are averaged. For some purposes such a procedure is suitable; but more often it is time-consuming, inaccurate, and even impossible when more than a few locations are to be sampled. Integrating-type light meters, which measure and integrate light for a full daylight period, are excellent instruments; but for most uses their cost is prohibitive.

In 1958, W. G. Dore briefly described a chemical light meter[1] that in several respects seemed superior to other light-measuring devices for use in silvical or ecological studies. It offers these advantages:

● It measures the cumulative amount of light reaching a particular location during a period of time.

● It is comparatively inexpensive.

● The number of locations that can be sampled simultaneously is essentially unlimited.

● The measurement can be made in almost any location where a small vial of the light-sensitive chemical can be placed.

The chemical light meter appears to have a wide range of application in forest research. It has undergone a year of testing at the Bartlett Experimental Forest in New Hampshire. It was used successfully during 1960 to obtain light measurements in a study of paper birch regeneration. Preliminary tests are also under way for investigating possible uses in other studies. Information from some of these tests is included in this report to illustrate how the method may be used.

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[1] Dore, W. G. A simple chemical light meter. Ecology 39:151-152, illus. 1958.

Although Dore's article outlined general procedures for using the chemical light meter, experience quickly revealed that anyone unfamiliar with the chemicals and the required operations still faces considerable difficulties in learning how to use it. To lessen those difficulties for other prospective users, detailed step-by-step instructions for manipulating the chemicals and converting the raw data obtained from them into standard units of light measurement are presented in this paper.

Principle of the Method

Anthracene $(C_{14}H_{10})$ in benzene will polymerize into insoluble dianthracene $(C_{14}H_{10})_2$ upon exposure to light. This property can be used to measure the amount of light that enters an environment over a period of time. In use, vials of the anthracene-benzene solution are exposed to the light of a particular environment for a period of time, then are analyzed to determine the amount of unconverted anthracene remaining in solution. This analysis is made with a spectrophotometer.[2] A standardization curve (relating percent transmittance from spectrophotometer to concentration of anthracene) and a calibration curve (relating concentration of anthracene to light exposure) are required for converting the chemical reaction into standard units of light.

Procedure

Determining a suitable wavelength to use in the spectrophotometer, and preparing standardization and calibration curves, are prerequisites to actual use of the method. However, the procedure may be more easily understood if the steps in making a light measurement are described first, and the fine details on determining the wavelength and preparing the curves are filled in later. The following are the steps involved in making a typical light measurement.

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[2] Spectrophotometer — an instrument used to measure the amount of light of a particular wavelength transmitted through the anthracene-benzene solution. The amount of light transmitted through the solution is related directly to the concentration of anthracene in the solution.

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Preparation
of Light-Sensitive Solution

Weigh out 6 grams ·of anthracene, using a balance that is accurate to 0.01 gram or less. Measure out 1 liter of benzene in a graduated cylinder. Dissolve the anthracene in the benzene in a flask or other convenient vessel (fig. 1).

This concentration (6 grams/liter) was selected for several reasons. It is a sufficiently high concentration to allow 7 to 8 hours' exposure in full sunlight. Higher concentrations are possible, but ordinarily are not needed; and it becomes difficult to dissolve the anthracene at concentrations higher than 6 g./l. Lower concentration would not allow a full day's exposure in full sunlight but might be desirable when the solutions are to be exposed in dense shade. We have used a concentration of 3 g./l. for these situations.
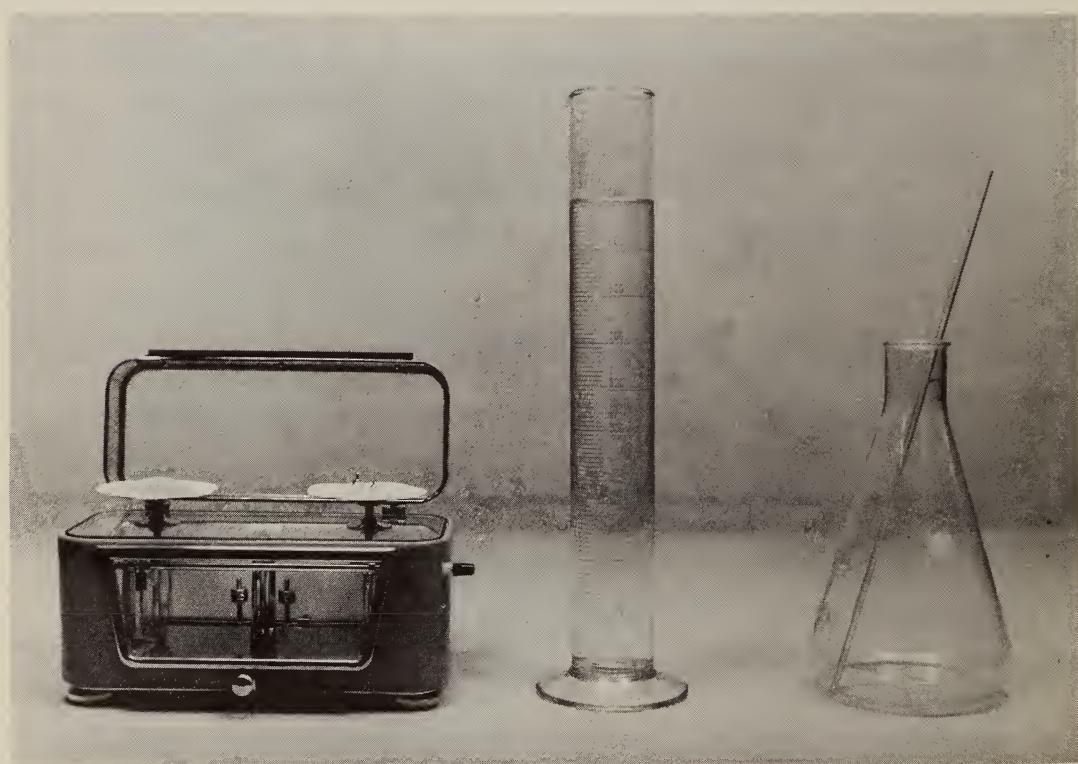
Figure 1. — First step: preparing the light-sensitive solution. An accurate balance is required.

Figure 2.—Filtering the solution to remove impurities.

Filter the solution (to remove any undissolved impurities) into a container large enough to hold all of the solution required for a given test (fig. 2).

The solution will not be affected by exposure to ordinary room light for short periods of time. However, exposure should be kept to a minimum. A dark bottle helps reduce possibility of too much exposure.

A sample of the stock solution should be tested in the spectro-photometer (as outlined later) to insure that the concentration is actually the desired 6 g./l.

Figure 3.—Filling vials with the chemical solution. A lazy susan device facilitates this operation.

Fill a supply of 50-milliliter vials from the stock bottle (fig. 3). Cap the vials and place them in a light-tight box. Vials should be filled as full as possible to reduce enclosed air space. One liter of solution fills 20 vials.

The bottle containing the stock solution is fitted with tubing, stopcock, and pressure bladder for easy filling of the vials. A lazy susan for holding the vials simplifies the job and permits two men to use production-line technique.

Caps must be foil-lined to protect against benzene, and even then they must be replaced after several uses.

Vials may be stored in the light-tight box for several days before exposure, if necessary. Prolonged storage periods should be avoided.

Light Measurement

Place the vials on the spot where you desire to make the light measurement. Small canvas pads 18 x 12 inches may be placed at the measurement spots to provide a standard background material and to insure that the same locations are used in subsequent measurements. The pads can be fastened to the ground with nails (fig. 4).
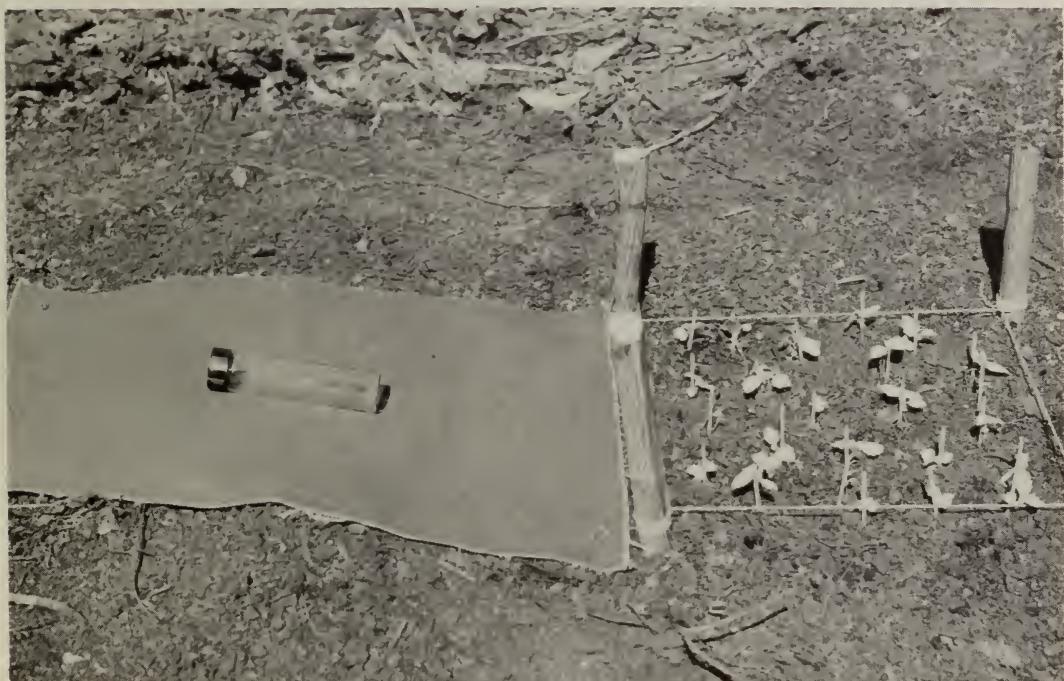

Figure 4.—Exposing a vial at a spot where light is to be measured.

Vials can be exposed to full sunlight for 6 to 8 hours under most conditions. When a series of micro-environments are being compared, and exposure times are short (1 day or less), vials should be picked up in the same sequence as laid out so that all are subjected to essentially the same exposure time. Also, for uniformity in relation to the sun's position, all vials should be oriented in the same direction.
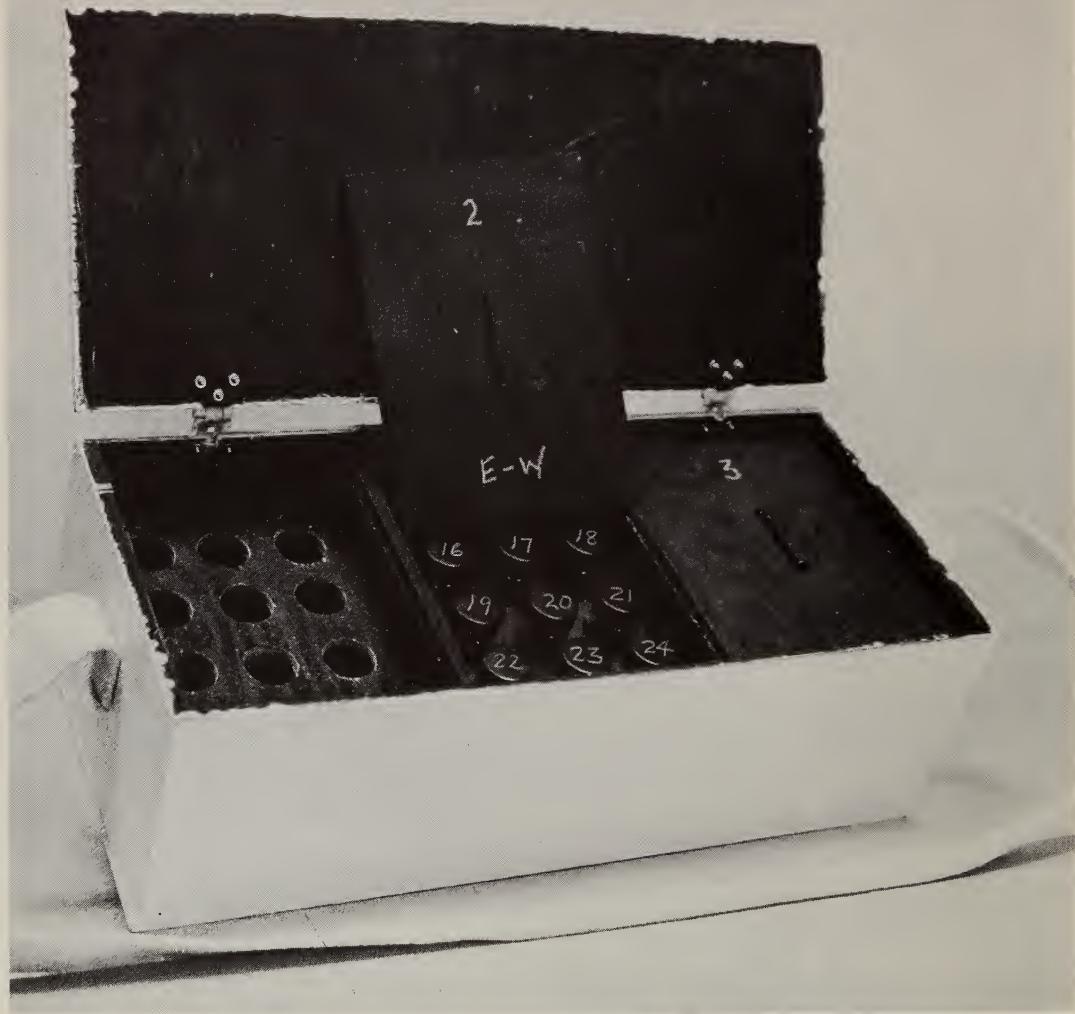
Figure 5. — The lightproof box devised for storing exposed vials.

During exposure, crystals of dianthracene will form and settle out of the solution.

After exposure, place the vials back in the light-tight box until they can be analyzed.

Exposed vials stored in these boxes were tested and were found unchanged after 4 days' storage. They could undoubtedly be stored longer, but this should be avoided. These plywood boxes were specially made for the purpose (fig. 5).

Analysis of Exposed Vials

First, filter the exposed solution into a clean vial (fig. 6). Since the analysis is made on the amount of anthracene remaining in solution rather than the amount of dianthracene formed, the dianthracene crystals must be removed. Most of the crystals will settle to the bottom of the exposed vial. Only about half of the exposed solution need be filtered; the other half, containing most of the dianthracene crystals, may be discarded.

For this, too, the lazy susan facilitates efficient handling of a large number of samples by a 2-man team. A lazy susan like this one can be made from plywood and wooden dowels, using a heavy ring stand as a base and pivot.
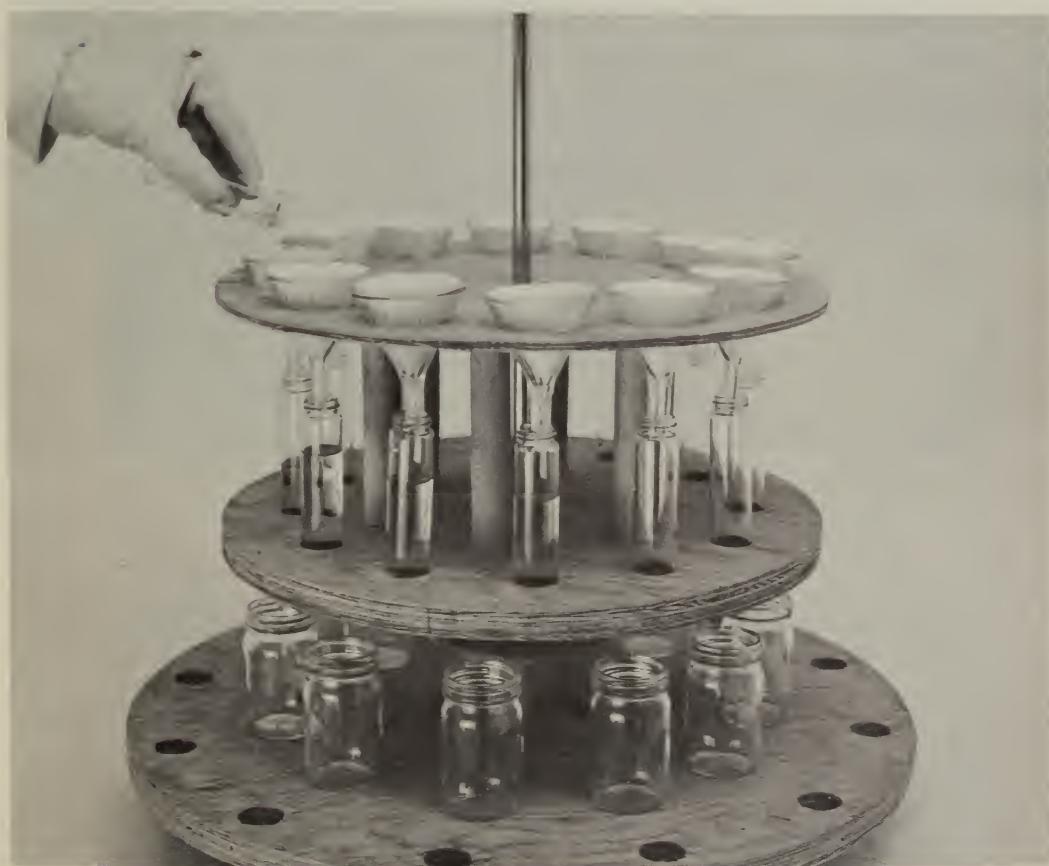
Figure 6.—First step in the analysis is to filter out the dianthracene crystals that have formed.
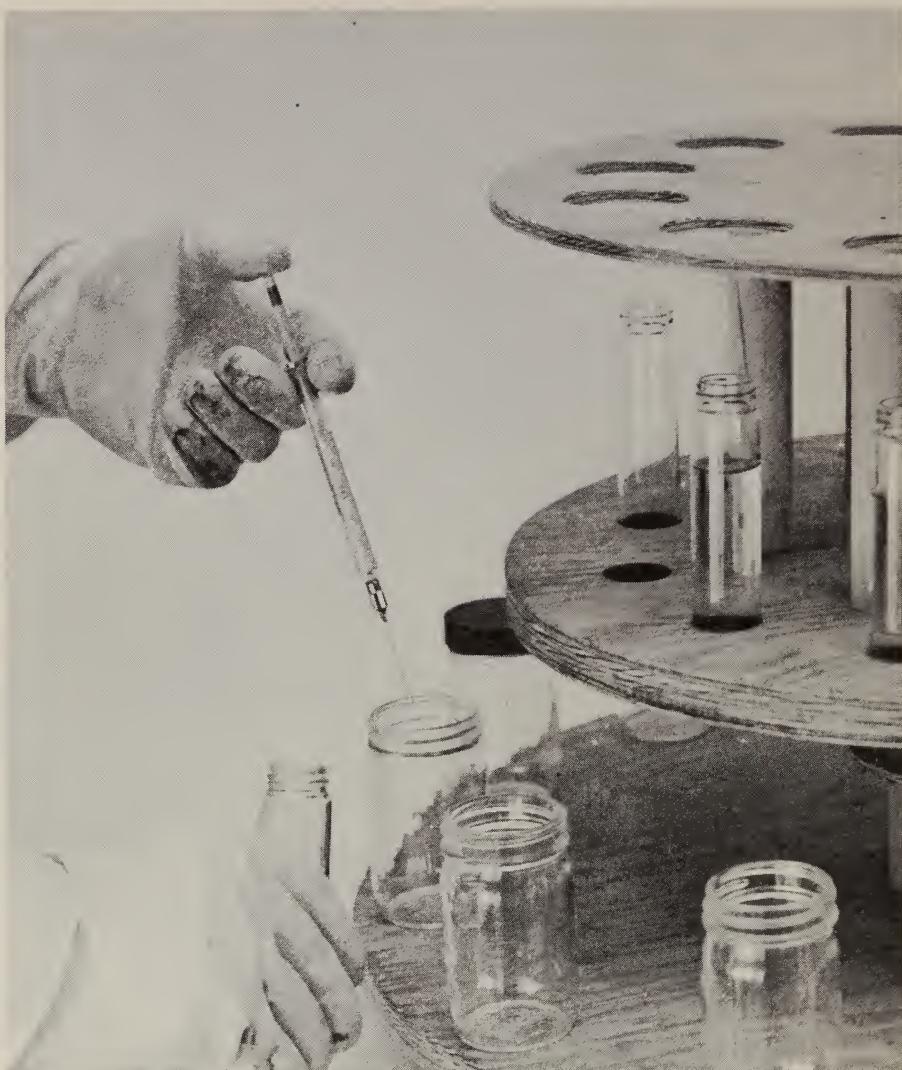
Figure 7. — An accurate syringe or micropipette is
required for taking the 0.5-ml. sample for analysis.

Then take a 0.5-ml. sample from the filtered solution and place
it in a clean 4 oz. jar.

The small quantity of solution that must be taken (0.5 ml.)
requires an accurate micropipette or syringe (fig. 7). The syringe
pictured above, which worked very well, was obtained from a
local drug store. Any instrument that requires mouth suction, or
that will not deliver the required quantity accurately and con-
sistently, is not suitable.

Figure 8. — Pure benzene is added to bring the test sample within the range of the spectrophotometer.

Add enough pure benzene to the 0.5-ml. sample to obtain a concentration within the range of the spectrophotometer (fig. 8). Record the dilution. The amount of benzene that must be added will vary from none to several hundred ml. Trial and error are the only guides to the amount of benzene required. As a starting point, try adding 30 ml., then test in the spectrophotometer. If a suitable reading is not obtained, make a new dilution. (See next step.)

For convenience of analysis, 10 to 30 ml. seems to be the best dilution. If dilutions of less than 5 ml. or more than 50 ml. are required, either the exposure time or the starting concentration should be changed.

A reservoir burette simplifies the job of diluting the samples.

Now cap the jar, mix the contents vigorously, and pour the solution into one of the special test tubes supplied for the spectrophotometer. Insert the test tube containing the sample into the cell of the spectrophotometer (fig. 9), which is set at the predetermined wavelength — in our work 350 millimicrons (mμ). You will obtain a reading in percent transmittance.
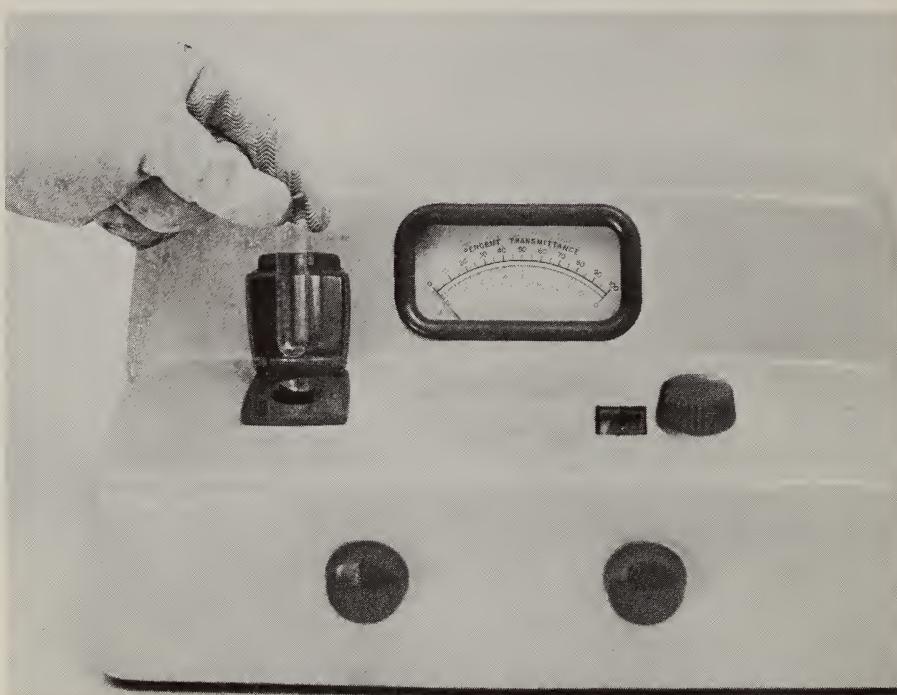

Figure 9. — Inserted into the spectrophotometer, the sample in the test tube produces a reading in percent of light transmitted.

The most accurate range of the instrument lies between 25 and 65 percent transmittance. If a sample is beyond these limits, another dilution must be made to bring it within the desired range.

Normal operating procedures, as outlined in the instruction manual for the spectrophotometer, must be followed. The standardization of the instrument should be checked frequently.

Using the percent transmittance obtained from the spectrophotometer, determine the corresponding concentration of anthra-

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cene (in mg./l.) from the standardization curve (fig. 10). This figure is the concentration of the diluted solution. Now compute the concentration of the solution before diluting, as follows:

Assume that you use 0.5 ml. of undiluted solution, and add 29.5 ml. of pure benzene to obtain 30 ml. of diluted solution. Testing the diluted solution in the spectrophotometer, you obtain a reading of 40 percent transmittance. From the standardization curve, you determine that this corresponds to a concentration of 13 mg./l.

13 mg./l. x .030 l. = 0.39 mg. anthracene in the diluted solution.
(30 ml.)
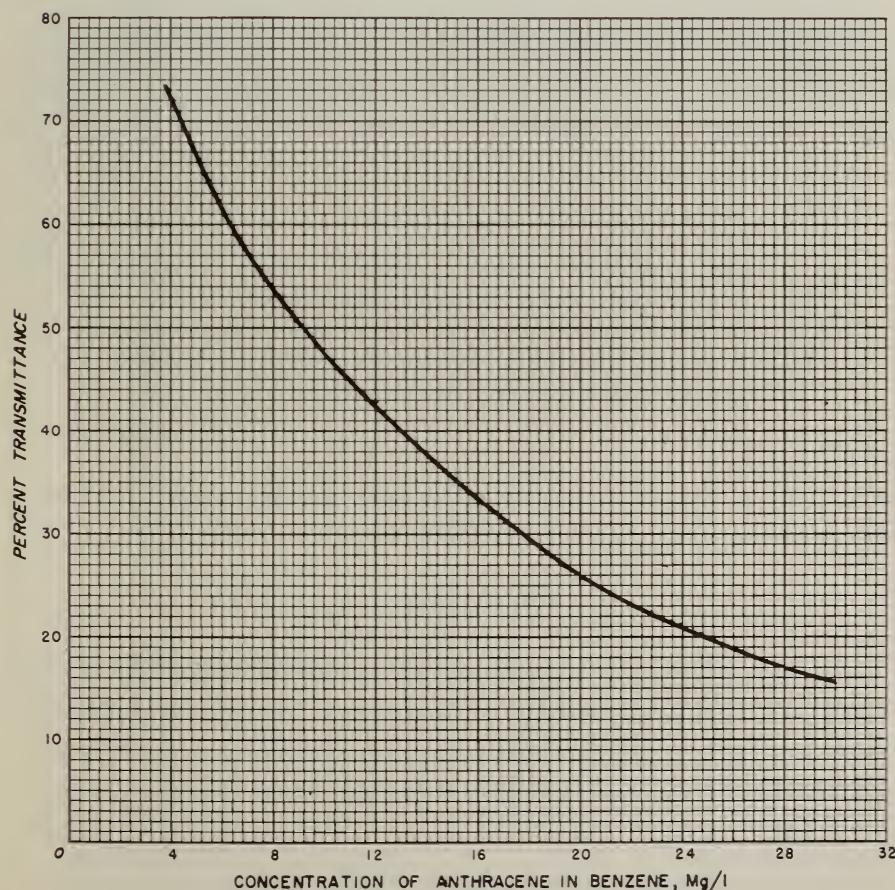
Figure 10.—The standardization curve. This relates concentration of solution to light transmittance as recorded by spectrophotometer.

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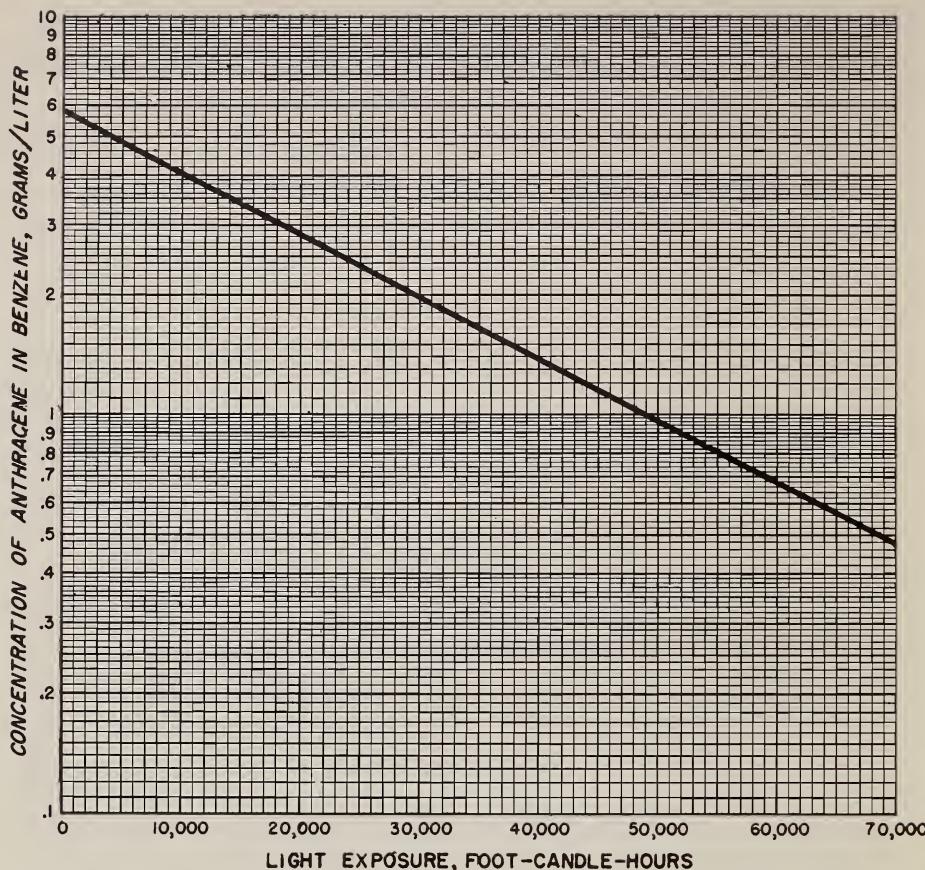
Figure 11. — The calibration curve, used to convert solution concentration data to foot-candle-hours.

Since all of this 0.39 mg. was present in the original 0.5-ml. sample, the concentration was:

$$\frac{0.39}{0.5} = 0.78 \text{ g./l.}$$

This computation can be reduced to a simple operation if the sample taken is held constant — in this case, 0.5 ml. Multiply the concentration obtained from the standardization curve by 1/5 volume of the diluted solution and move the decimal point 2 places to the left. Thus:

$$13 \text{ x } 6 = 78 = 0.78$$
$$(1/5 \text{ x } 30)$$

Standardizing dilutions at a figure easily divided by 5 also simplifies calculations.

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After computing the concentration of the undiluted solution, determine the light exposure from the calibration curve (fig. 11). This figure, in foot-candle-hours, can be used for comparison with other vials exposed during the same period. In the example above, 0.78 g./l. corresponds to an exposure of 55,500 foot-candle-hours.

Wavelength Determination

Since the wavelength of light used in the spectrophotometer affects the transmittance readings, one wavelength must be se- lected and used for all tests. In our work a wavelength of 350 mμ was used. This was selected by preparing curves of percent trans- mittance over wavelength. Separate curves were prepared for each of several concentrations. The exact concentration is not critical because all curves will be similar in shape. It is important only that the concentration used give readings over the middle range of percent transmittance (25 to 65 percent). One of these curves,
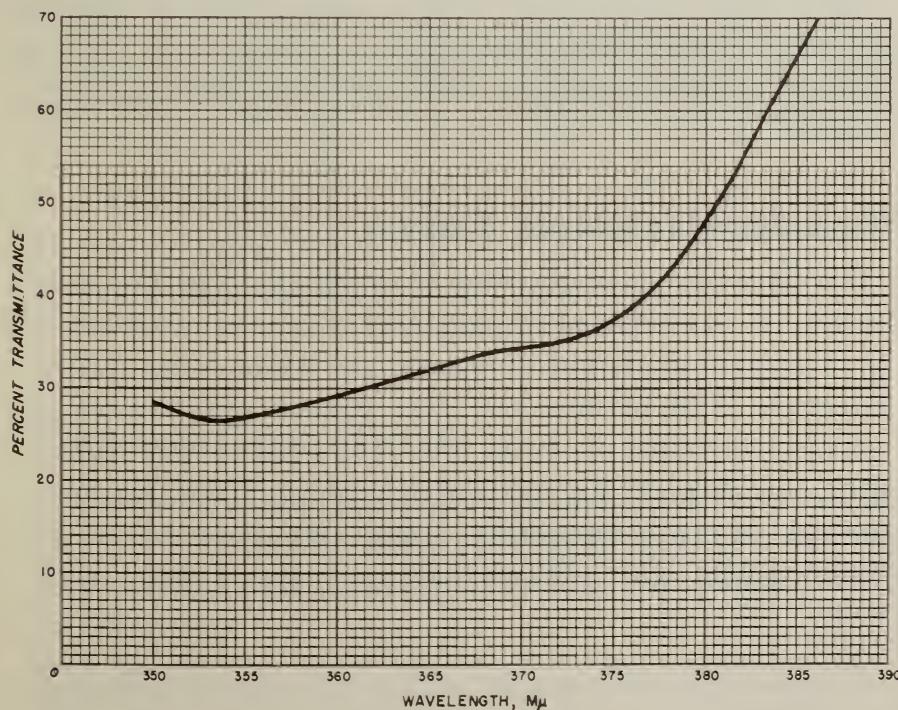

Figure 12.—A curve for wavelength determination.

for a concentration of 20 mg./l. is shown in figure 12. The wave-length was selected from this curve on the following criteria:

● A wavelength should be selected that absorbs a maximum amount of light (and thus transmits a minimum), because this analysis is essentially absorption measurement.

● A point on a steeply rising or falling section of the curve should be avoided because a small wavelength error could cause a large transmittance error.

● The concentrations that it is desired to test should fall in the 25 to 65 percent transmittance range because this is the most accurate range of the instrument.

As evidenced from the curve in figure 12, a wavelength between 350 and 360 mμ would satisfy the first and second criteria. But no wavelength will allow field solutions (which have a concentration of from 6 g./l. to about 0.5 g./l.) to be tested in the 25 to 65 percent range. Therefore the field solutions must be diluted to bring them within the range.

For practical purposes, 350, 355, or 360 mμ would all have been equally satisfactory wavelengths. Of these three, 350 mμ was chosen simply to conform to the wavelength used by Dore in his work.[3]

Standardization Curve

This curve relates concentration of anthracene to percent transmittance. It was obtained by preparing a solution of about 30 mg. of anthracene per liter of benzene and testing in the spectrophotometer. This solution was then diluted with benzene to obtain concentrations of 28, 26, 24 . . . 4, 2 mg./l. Each concentration was tested in the spectrophotometer to obtain the corresponding percent transmittance. These data were plotted on ordinary graph paper with percent transmittance over concentration.

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[3] Different spectrophotometers may give slightly different results. We used a Spectronic 20, manufactured by Bausch and Lomb. Dore also used this type of instrument. In correspondence with Dore, however, he indicated that one of his colleagues (D. C. J. Friend) had obtained two peak absorbencies in some trials he made with a Beckman spectrophotometer. These two peak absorbencies were at 360 and 379 mμ.

A standardization table was made from this curve because it is slightly easier to read than the curve itself. This is table 1.

Table 1.—Standardization table.

Percent transmittance	mg./l.	Percent transmittance	mg./l.	Percent transmittance	mg./l.
25	20.6	39	13.5	53	8.5
	20.3		13.3		8.3
26	20.0	40	13.1	54	8.2
	19.6		12.9		8.0
27	19.2	41	12.7	55	7.9
	18.9		12.5		7.7
28	18.6	42	12.3	56	7.6
	18.4		12.1		7.4
29	18.0	43	12.0	57	7.3
	17.8		11.8		7.2
30	17.5	44	11.6	58	7.0
	17.3		11.4		6.9
31	17.0	45	11.2	59	6.8
	16.7		11.0		6.6
32	16.4	46	10.8	60	6.5
	16.2		10.6		6.4
33	16.0	47	10.4	61	6.2
	15.8		10.3		6.1
34	15.6	48	10.1	62	6.0
	15.3		10.0		5.9
35	15.1	49	9.8	63	5.8
	14.9		9.6		5.6
36	14.7	50	9.4	64	5.5
	14.5		9.3		5.4
37	14.3	51	9.1	65	5.3
	14.1		9.0		
38	13.9	52	8.8		
	13.7		8.6		

Calibration Curve

This curve relates concentration of anthracene to foot-candle-hours. It is devised by comparing exposed anthracene vials with some standard light-measuring instrument. The procedure was as follows:

- Screens were constructed to provide four degrees of shade. These, plus full sunlight, provided five light intensities. Con-

Figure 13.—Set-up for calibration. The four screens and open table top provide five different light intensities. The larger black instrument is the illumination meter, with target attached; the smaller is a clock for timing intervals between exposures.

structed of saran shade screening and/or cheesecloth, supported on pieces of 1 x 2-inch lumber, the screens were placed on a table top that was covered with a standard background material — canvas in this case (fig. 13).

The relative value of each light intensity was determined by a series of twenty readings with a Weston Illumination meter. Relative intensity values (RI) were: 1, 0.85, 0.67, 0.54, and 0.37.

• Twenty vials were filled with a stock solution of 6 g./l., and four vials were exposed under each of the five light intensities. One vial was removed from each intensity after 3 hours' exposure, one after 4 hours, one after 5, one after 6.

During the entire 6-hour period, measurements of light intensity were taken with the illumination meter at 5-minute intervals under one of the screens. These measurements were averaged to obtain the intensity for each hour of the 6-hour period. Intensities under the other screens and under full sunlight were computed from this data and the RI values. The intensity-duration figure was then computed by summing the intensities of the first 3 hours, the first 4 hours, the first 5 hours, and the entire 6 hours.

After exposure, the vials were analyzed with the spectrophotometer and the concentration of anthracene remaining in solution was plotted over the appropriate light intensity-duration (in foot-candle-hours). Semilog paper was used for this because the conversion falls off according to a logarithmic curve as the solution became weaker in anthracene, thus forming a straight line on semilog paper (fig. 11).

The straight line was fitted to the data by regression correlation. A correlation coefficient of 0.99 was obtained. Some of the calibration data are shown in table 2.

Table 2.—Foot-candle-hours in full sunlight for periods of 3, 4, 5, and 6 hours, as computed from illumination meter readings during the calibration, and concentrations of anthracene after exposure for the different periods under the different light intensities.

(Starting concentration — 6 g./l.)

Time period (hours)	Intensity-duration under full sunlight	Screen No. 1 (RI = .370)	Screen No. 2 (RI = .543)	Screen No. 3 (RI = .674)	Screen No. 4 (RI = .845)	Full Sunlight (RI = 1.000)
	Foot-candle hours	g./l.	g./l.	g./l.	g./l.	g./l.
3	24800	4.38	3.48	3.18	2.52	2.35
4	35300	3.78	2.94	2.50	1.72	1.58
5	45400	3.30	2.40	1.96	1.39	1.20
6	54200	2.98	2.08	1.60	1.06	.86

Intensity of
Full Sunlight

The day chosen for calibration should be a nearly cloudless one. Such a day is ideal because danger of error from rapid-changing intensities common on partly cloudy days is avoided. The pattern of light intensity during such a cloudless day is shown in fig. 14.
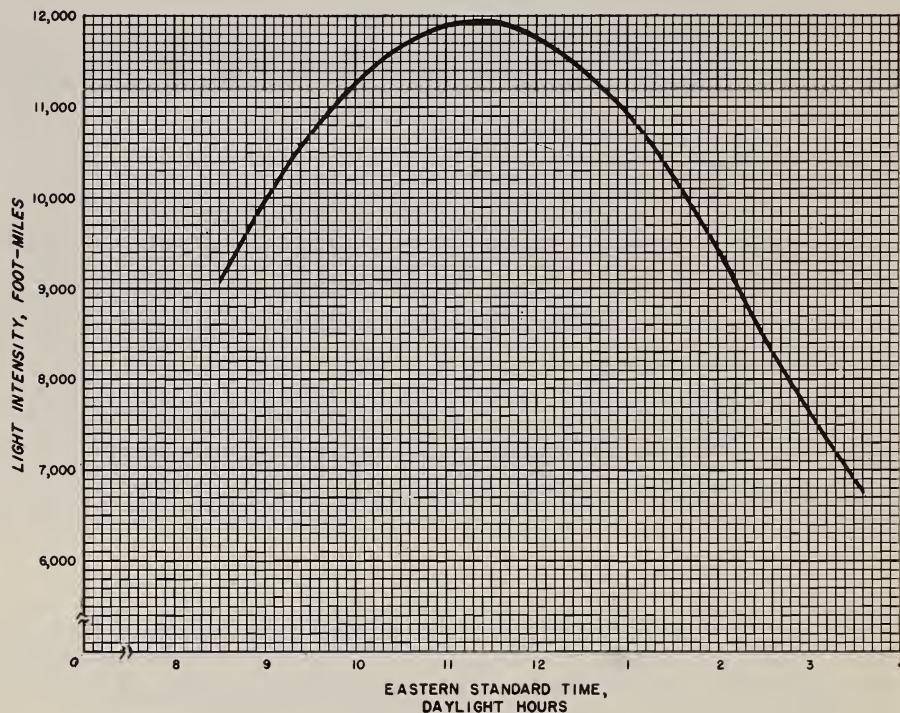

Figure 14.—Pattern of light intensity for a cloudless day.

Laboratory Techniques

General Considerations

A well-equipped laboratory is convenient, but by no means essential, for preparation and analysis of the anthracene solutions. Our work was done in an ordinary kitchen. Basic requirements of the workroom are simple: running water, adequate counter or table space, storage facilities, and some means for venting the benzene fumes.

Regardless of the place, techniques and procedures must conform to top laboratory standards — clean glassware, accurate measurements, thorough mixing, and proper care and use of instruments. Careless work almost certainly will yield faulty results.

It is important to recognize that the standardization and calibration data and curves may vary with equipment and purity of chemicals. Therefore, the same techniques and materials that will be used in the field measurements should be used for constructing the curves. Further, it is important that exactly the same techniques, the same stocks of chemicals, the same instruments, and the same sizes and types of critical glassware (such as the exposure vials) be used throughout any series of measurements.

Safety Precautions

Both materials required by this method must be handled with care. Benzene is highly flammable; it is posionous if swallowed, and the fumes may cause headache. Anthracene may cause skin irritations. However, there is little danger if a few simple safety precautions are observed:

• Some type of ventilation, such as an exhaust fan, must be provided in the room where analysis is to be made. Large fans mounted in the window are ideal in rooms not properly equipped.

• NO SMOKING signs should be posted and should be strictly enforced.

- Benzene should be stored in a safe place, in a safe container.
- Tight-fitting rubber gloves should be worn when handling solutions, and hands should be thoroughly washed with soap and water after completion of the work.

In case of accident, treat as follows:

- If solutions come in contact with skin: wash in soap and water.
- If solutions are swallowed: induce vomiting by swallowing hot, soapy water. Call doctor.
- If solution gets into the eyes: flush immediately with water.
- If fumes cause unconsciousness or stop breathing: call doctor, get victim into fresh air, give artificial respiration.

Chemical
Light Meter in Use

The following examples illustrate some of the uses of the chemical light meter.

Regeneration Studies

In a study of birch regeneration, 78 seedspots were located at various positions within clearcut patches of about ¾-acre size. The amount of light reaching each seedspot varied with its location in relation to border trees. The objective was to determine the effect of various amounts of light on seedling growth.

The chemical light meter was used to measure light on each of the 78 seedspots. In the graph below, the height growth of

HEIGHT

LIGHT EXPOSURE
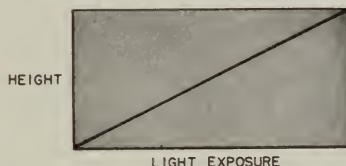
1-year-old paper birch seedlings is plotted over amount of light reaching each seedspot over a 6-hour period. The curve was fitted by regression correlation. The correlation coefficient proved significant at the 0.001 level.

Stand or Crown Density Studies

To evaluate the chemical light meter as an instrument for measuring light under dense forest canopies, vials of anthracene-benzene were exposed under stands that had been cut to various basal-area specifications. Technique here differed from that used in open areas. A concentration of 3 g./l. was used and the vials were exposed for 1 week rather than 1 day.

In one study, 20 vials were exposed in each of two ¼-acre plots. One plot had 40 percent of its basal area removed the previous year. The other plot was uncut. The average amount of light

reaching the forest floor under the uncut stand was 6,375 foot-candle-hours versus 10,370 foot-candle-hours under the cut stand. These differences were highly significant.

The purpose of this test was to determine whether the method can be used under forest canopies where light intensity is very low. Further tests are needed, and recalibration will be necessary for use under such low intensities. However, it seems probable that the chemical light meter can be used to advantage in this type of study.

The above examples are included merely to suggest some uses of the chemical light meter. There are undoubtedly many other types of studies to which the method could be applied.

Equipment Required

The following list of equipment is presented in considerable detail so that other users can closely duplicate the results shown in this report. Most of the minor items on the list are standard laboratory equipment. The specifications given for many of these articles need not be rigid; at a working laboratory suitably similar if not identical glassware, stoppers, supports, and the like would be on hand. The main condition to be imposed here is that exactly the same size and type of such critical items as exposure vials be available in adequate supply.

1	Spectrophotometer, Bausch & Lomb Spectronic 20. Colorimeter or equivalent.
1	Balance (of suitable sensitivity — to 0.01 gram).
1	Illumination meter, Weston Model 756 or equivalent.[4]
2.5X	Vials, screw top, 108 x 28 mm.
2.5X	Caps, foil-lined, size 24.
X	Bottles, wide-mouth, screw-cap, 4-ounce.
X	Caps, foil-lined, size 48.
X	Funnels, glass, $2\frac{5}{8}$-inch diameter, $\frac{1}{8}$ pint.
1-2	Funnels, glass, 7-inch diameter, 1 quart.
As needed	Benzene.
As needed	Filter paper, Whatman #2, 125 mm.

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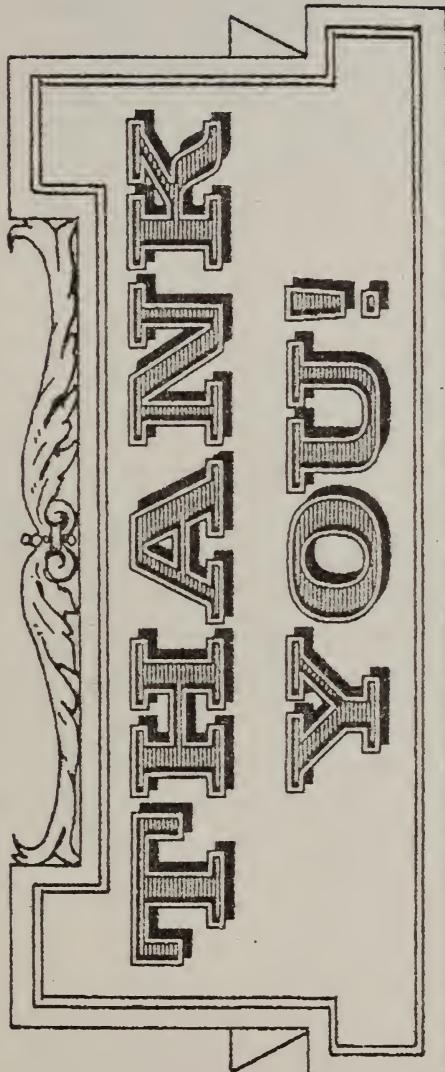
As needed	Filter paper, Whatman #2, 270 mm.
As needed	Anthracene.
1	Micropipette.[5]
1	Burette, reservoir.
X	Test tubes to fit the spectrophotometer.
Several	Graduated cylinders, 1000 ml.
Several	Graduated cylinders, 250 ml.
Several	Graduated cylinders, 25 ml.
	Rubber stoppers, 2-hole, size 4.
	Glass tubing, plastic tubing, glass stopcock, etc.
	Test tube supports.
	Wash baskets.
	Ring stand.
	Stirring rods, 15-inch, $3/8$-inch diameter.
	Light-tight vial box (homemade).
	Lazy susan funnel and vial rack (homemade).

Nearly all the articles on the list can be obtained from laboratory supply companies. The required number of some items is indicated. "X" in the list denotes the number of points to be sampled or vials to be exposed simultaneously. Much time can be saved by having enough equipment to permit a complete run of a series without stopping to wash glassware.

The most expensive piece of equipment is the spectrophotometer (around $300). Where most of the standard laboratory equipment is on hand, and an illumination meter is available, the cost of the spectrophotometer will be the only substantial cash outlay required.

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[4] Illumination meter is needed only for the calibration.

[5] Micropipette — this is the weak point in the system because of the small quantity (0.2 to 0.5 ml.) which must be measured accurately. *A good pipette or syringe is essential.* A tuberculin syringe, graduated in 1/100 ml., and fitted with an attachment that automatically dispenses a preset quantity, was used in our studies.